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Search Results -

Terms	Documents
(uemura or okui or kominami or yamaguchi or mitsui).in. and (vecto$3 or plasmid$3) and (tag$3 same cleav$5 same secret$5)	1

Database: US Patents Full-Text Database; US Pre-Grant Publication Full-Text Database; JPO Abstracts Database; EPO Abstracts Database; Derwent World Patents Index; IBM Technical Disclosure Bulletins

Search: L2

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Search History

DATE: Thursday, October 30, 2003 Printable Copy Create Case

Set Name side by side	Query	Hit Count	Set Name result set
DB=USPT,PGPB,JPAB,EPAB,DWPI; PLUR=YES; OP=OR			
L2	(uemura or okui or kominami or yamaguchi or mitsui).in. and (vecto$3 or plasmid$3) and (tag$3 same cleav$5 same secret$5)	1	L2
L1	secret$5 same tag$3 same (vecto$3 or plasmi$3) same cleav$5 same space$3	6	L1

END OF SEARCH HISTORY

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Search Results - Record(s) 1 through 6 of 6 returned.

1. Document ID: US 20030166160 A1

L1: Entry 1 of 6 File: PGPB Sep 4, 2003

PGPUB-DOCUMENT-NUMBER: 20030166160
PGPUB-FILING-TYPE: new
DOCUMENT-IDENTIFIER: US 20030166160 A1

TITLE: Compounds and molecular complexes comprising multiple binding regions directed to transcytotic ligands

PUBLICATION-DATE: September 4, 2003

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Hawley, Stephen B.	San Diego	CA	US	
Chapin, Steven	San Diego	CA	US	
Sheridan, Philip L.	San Diego	CA	US	
Houston, L. L.	Del Mar	CA	US	
Glynn, Jacqueline M.	San Diego	CA	US	

US-CL-CURRENT: 435/69.7; 435/320.1, 435/325, 435/6, 530/350, 536/23.5

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | Claims | KWIC | Draw Desc | Image

2. Document ID: US 20030161809 A1

L1: Entry 2 of 6 File: PGPB Aug 28, 2003

PGPUB-DOCUMENT-NUMBER: 20030161809
PGPUB-FILING-TYPE: new
DOCUMENT-IDENTIFIER: US 20030161809 A1

TITLE: Compositions and methods for the transport of biologically active agents across cellular barriers

PUBLICATION-DATE: August 28, 2003

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Houston, L. L.	Del Mar	CA	US	
Sheridan, Philip J.	San Diego	CA	US	
Hawley, Stephen B.	San Diego	CA	US	
Glynn, Jacqueline M.	San Diego	CA	US	
Chapin, Steven	San Diego	CA	US	

US-CL-CURRENT: 424/85.2; 424/178.1, 435/6, 514/44, 530/351, 530/391.1, 530/395

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | Claims | KWIC | Draw Desc | Image

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3. Document ID: US 20020160482 A1

L1: Entry 3 of 6 File: PGPB Oct 31, 2002

PGPUB-DOCUMENT-NUMBER: 20020160482
PGPUB-FILING-TYPE: new
DOCUMENT-IDENTIFIER: US 20020160482 A1

TITLE: Methods for protein purification

PUBLICATION-DATE: October 31, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Abrahmsen, Lars	Bromma		SE	
Nilsson, Joakim	Danderyd		SE	

US-CL-CURRENT: 435/191; 435/320.1, 435/325, 435/69.1, 536/23.2

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | Claims | KWIC | Draw Desc | Image

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4. Document ID: US 20020009455 A1

L1: Entry 4 of 6 File: PGPB Jan 24, 2002

PGPUB-DOCUMENT-NUMBER: 20020009455
PGPUB-FILING-TYPE: new
DOCUMENT-IDENTIFIER: US 20020009455 A1

TITLE: DNA encoding a novel PROST 03 polypeptide

PUBLICATION-DATE: January 24, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Lau, Ted	Alameda	CA	US	
Lin, Richard J.	Danville	CA	US	
Parkes, Deborah	Hayward	CA	US	
Parry, Gordon	Walnut Creek	CA	US	
Schneider, Douglas W.	Lafayette	CA	US	
Steinbrecher, Renate	Walnut Creek	CA	US	
Heuit, Pamela Toy Van	Moraga	CA	US	
Wu, John	Carlisle	MA	US	

US-CL-CURRENT: 424/178.1; 435/325, 435/6, 435/69.1, 435/7.23, 530/350

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc
Image

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5. Document ID: US 20020004047 A1

L1: Entry 5 of 6 File: PGPB Jan 10, 2002

PGPUB-DOCUMENT-NUMBER: 20020004047
PGPUB-FILING-TYPE: new
DOCUMENT-IDENTIFIER: US 20020004047 A1

TITLE: DNA encoding a novel RG1 polypeptide

PUBLICATION-DATE: January 10, 2002

INVENTOR-INFORMATION:

NAME	CITY	STATE	COUNTRY	RULE-47
Harkins, Richard	Alameda	CA	US	
Parkes, Deborah	Hayward	CA	US	
Parry, Gordon	Walnut Creek	CA	US	
Schneider, Douglas W.	Lafayette	CA	US	
Steinbrecher, Renate	Walnut Creek	CA	US	

US-CL-CURRENT: 424/178.1; 435/325, 435/6, 435/69.1, 435/7.23, 530/388.8

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc
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6. Document ID: US 5935824 A

L1: Entry 6 of 6 File: USPT Aug 10, 1999

US-PAT-NO: 5935824
DOCUMENT-IDENTIFIER: US 5935824 A

TITLE: Protein expression system

DATE-ISSUED: August 10, 1999

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Sgarlato; Gregory D.	Los Gatos	CA		

US-CL-CURRENT: 435/69.7; 435/69.8, 530/350, 536/23.4

Full | Title | Citation | Front | Review | Classification | Date | Reference | Sequences | Attachments | KWIC | Draw Desc
Image

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Terms	Documents
secret$5 same tag$3 same (vecto$3 or plasmi$3) same cleav$5 same space$3	6

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Search Results - Record(s) 1 through 1 of 1 returned.

1. Document ID: EP 1132479 A1

L2: Entry 1 of 1 File: EPAB Sep 12, 2001

PUB-NO: EP001132479A1
DOCUMENT-IDENTIFIER: EP 1132479 A1
TITLE: PROTEIN EXPRESSION VECTOR AND UTILIZATION THEREOF

PUBN-DATE: September 12, 2001

INVENTOR-INFORMATION:

NAME	COUNTRY
UEMURA, HIDETOSHI	JP
OKUI, AKIRA	JP
KOMINAMI, KATSUYA	JP
YAMAGUCHI, NOZOMI	JP
MITSUI, SHINICHI	JP

INT-CL (IPC): C12 N 15/86; C12 N 5/10; C12 P 21/02
EUR-CL (EPC): C12N009/64

Full Title CIT.1 REV.1 CLS.1 REF.1 SEQ.1 ATT.1 DRAW.1

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Terms	Documents
(uemura or okui or kominami or yamaguchi or mitsui).in. and (vecto$3 or plasmid$3) and (tag$3 same cleav$5 same secret$5)	1

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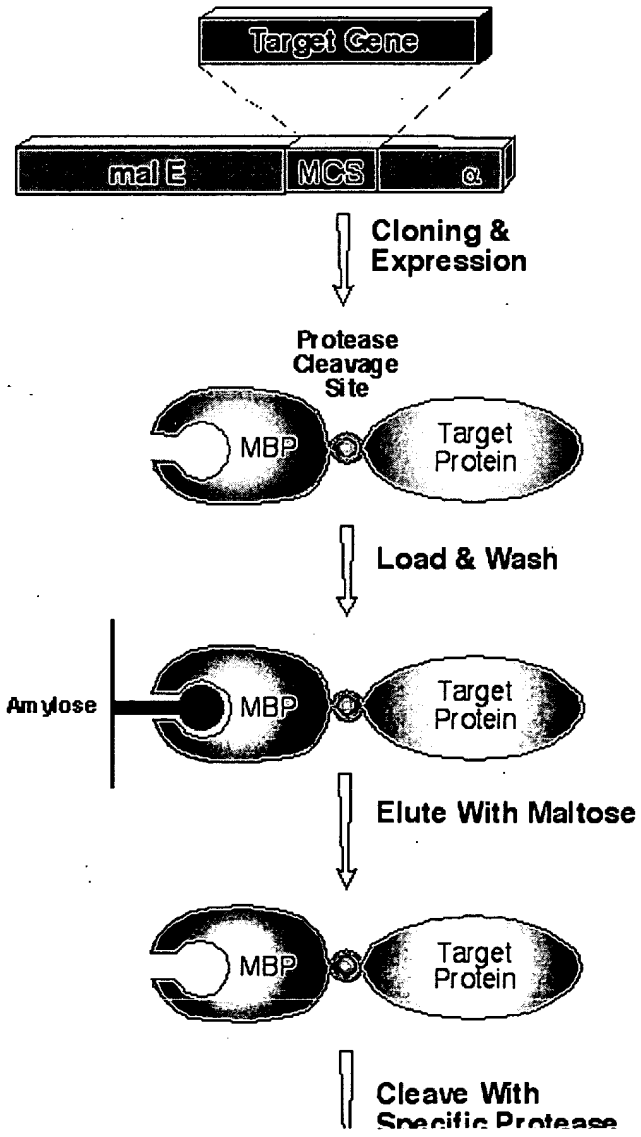
Protein Fusion & Purification (pMAL™) System

#E8000S **$400 (USA)**

Download: Technical Bulletin and Manual | Vector Sequence Files | FAQs |

Description: In the pMAL™ Protein Fusion and Purification System, the cloned gene is inserted into a pMAL vector down-stream from the malE gene, which encodes maltose-binding protein (MBP). This results in the expression of an MBP-fusion protein (1,2,3). The technique uses the strong P_{tac} promoter and the translation initiation signals of MBP to express large amounts of the fusion protein. The fusion protein is then purified by a one-step affinity purification specific for MBP (Figure 1) (4).



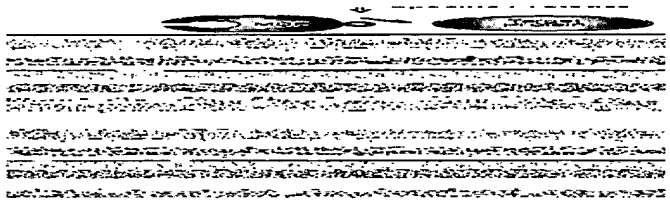


Figure 1: Schematic of the pMAL System.

The system uses the pMAL vectors which are designed so that insertion interrupts a *lacZ*a gene allowing a blue-to-white screen for inserts on X-gal (5). The vectors include a sequence coding for the recognition site of a specific protease. This allows the protein of interest to be cleaved from MBP after purification, without adding any vector-derived residues to the protein (6). For this purpose, the polylinker includes a restriction site superimposed on the sequence coding for the site of the specific protease. This is where the gene of interest is inserted.

Expression from the pMAL vectors yields up to 100 mg fusion protein from a liter of culture (Figure 2). In most cases, the expressed protein is soluble, as fusion to MBP has been proven to enhance the solubility of proteins expressed in *E. coli* (7). While no expression system works with every cloned gene, the pMAL Protein Fusion and Purification System gives substantial yields of protein in more than 75% of the cases tested. A chapter in *Current Protocols in Molecular Biology* (3) provides an in-depth analysis of the use of the pMAL vectors.

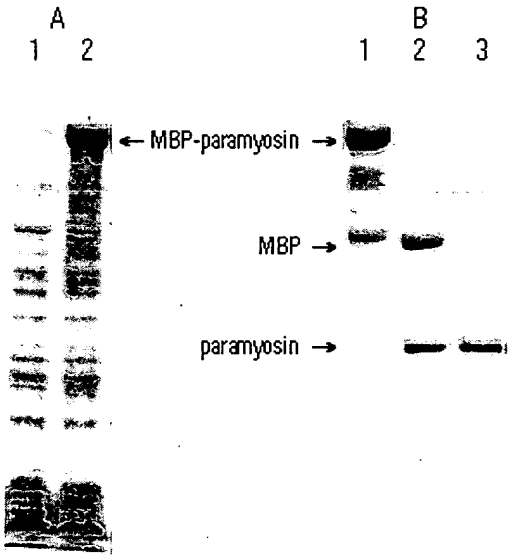


Figure 2: SDS-polyacrylamide gel electrophoresis of fractions from the purification of MBP-paramyosin-.Sal.
A: Lane 1: uninduced cells. Lane 2: induced cells.
B: Lane 1: purified protein eluted from amylose column with maltose. Lane 2: purified protein after Factor Xa cleavage. Lane 3: paramyosin fragment eluted from second amylose column.

The System Includes:

- pMAL-c2X: 10 µg

- pMAL-p2X: 10 µg
- Amylose Resin: 15 ml (binding capacity ~40 mg)
- Factor Xa: 50 µg
- Anti-MBP antiserum: 25 µl (for Western blot analysis)
- MBP2*: 10 µg (marker for SDS-polyacrylamide gels)
- MBP2*-paramyosin-DSal: 100 µg (control for Factor Xa cleavage)
- *E. coli* Host
- Comprehensive Instruction Manual

Advantages of the pMAL System:

- Reliable expression: substantial yields (up to 100 mg/L) in more than 75% of the cases tested.
- Expression in either the cytoplasm or periplasm: periplasmic expression enhances folding of proteins with disulfide bonds.
- Fusion to MBP has been shown to enhance the solubility of proteins expressed in *E. coli* (7).
- Gentle elution with maltose: no detergents or harsh denaturants.

References:

1. Guan, C. et al. (1987) *Gene* 67, 21–30.
2. Maina, C. V. et al. (1988) *Gene* 74, 365–373.
3. Riggs, P. D. (1990). In F. M. Ausebel, R. Brent, R. E. Kingston, D. D. Moore, J. G. Seidman, J. A. Smith and K. Struhl (Eds.), *Current Protocols in Molecular Biology* (pp. 16.6.1–16.6.10). New York: John Wiley & Sons, Inc.
4. Kellerman, O. K. and Ferenci, T. (1982). In W. A. Wood (Ed.), *Methods in Enzymology* Vol. 90, (pp. 459–463). New York: Academic Press.
5. Yanisch–Perron, C. et al. (1985) *Gene* 33, 103–119.
6. LaVallie, E. R. and McCoy, J. M. (1990). In F. M. Ausebel, R. Brent, R. E. Kingston, D. D. Moore, J. G. Seidman, J. A. Smith and K. Struhl (Eds.), *Current Protocols in Molecular Biology* (pp. 16.4.1–16.4.17). New York: John Wiley & Sons, Inc.
7. Kapust and Waugh (1999) *Protein Science* 8, 1668–1674.

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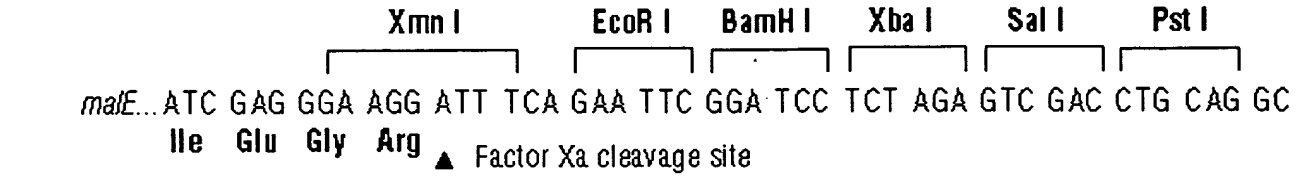
pMAL™ Companion Products

Vectors:

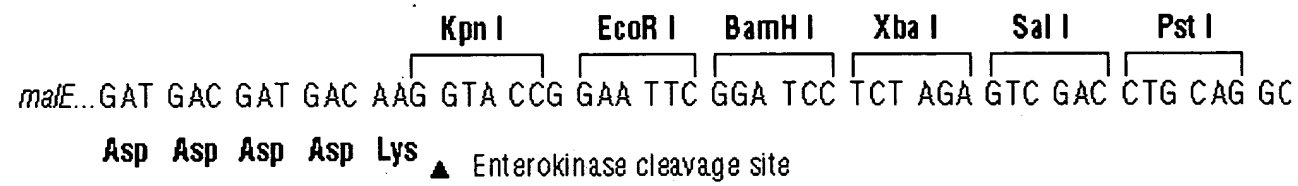
Description: The pMAL™-c2 series of vectors have an exact deletion of the malE signal sequence, resulting in cytoplasmic expression of the fusion protein. The pMAL-p2 series of vectors contain the normal *malE* signal sequence, which directs the fusion protein through the cytoplasmic membrane. All of the vectors include a sequence coding for the recognition site of a specific protease [Factor Xa (X), Enterokinase (E) or Genenase™ I (G)] which allows the protein of interest to be cleaved from MBP after purification.

---

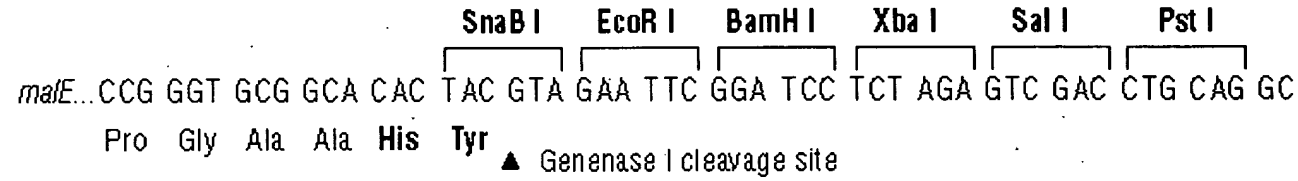
pMAL-c2X, pMAL-p2X Polylinker



pMAL-c2E, pMAL-p2E Polylinker



pMAL-c2G, pMAL-p2G Polylinker



N8076S	pMAL-c2X	10 µg	$75 (USA)
N8077S	pMAL-p2X	10 µg	$75 (USA)
N8066S	pMAL-c2E	10 µg	$75 (USA)
N8067S	pMAL-p2E	10 µg	$75 (USA)
N8068S	pMAL-c2G	10 µg	$75 (USA)
N8069S	pMAL-p2G	10 µg	$75 (USA)

Note: pMAL-c2X and pMAL-p2X are supplied with the pMAL Protein Fusion and Purification System (NEB #E8000S). Others are not supplied with the system.

Amylose resin:

Description: Affinity matrix used for isolation of proteins fused to maltose-binding protein. It is a composite amylose/agarose bead.

Binding Capacity: 2–4 mg MBP/ml bed volume.

Shipping: Supplied pre-swollen in 20% ethanol. Store at 4°C.

E8021S	15 ml	$100 (USA)
E8021L	100 ml	$300 (USA)

Factor Xa, Enterokinase and Genenase™ I :

Factor Xa

E8010S	50 µg	$50 (USA)
E8010L	250 µg.	$200 (USA)

Enterokinase (*not supplied with the system*)

E8070S	5 units	$80 (USA)
E8070L	25 units	$320 (USA)

Genenase™ I (*not supplied with the system*)

E8075S	50 µg	$50 (USA)
E8075L	250 µg	$200 (USA)

Anti-MBP serum:

Description: Rabbit antiserum prepared by immunizing with affinity-purified maltose-binding protein.

Concentration and Shipping: End-point dilution titer by Elisa greater than 10–4. Supplied frozen. Store at –20°C.

E8030S	0.2 ml	$100 (USA)

Anti-MBP Monoclonal Antibody: (*not supplied with the system*)

Description: Anti-MBP Monoclonal Antibody is purified from tissue culture fluid of hybridoma cell line B48 by protein A affinity chromatography

Concentration and Shipping: 1 mg/ml. Supplied in 10 mM HEPES (pH 7.5), 150 mM NaCl and 50% glycerol. Store at –20°C.

E8032S	0.05 ml	$125 (USA)
E8032L	0.25 ml	$500 (USA)

MBP2*-paramyosin DSal:

Description: MBP2*-paramyosin-DSal is a maltose binding fusion protein (MBP) containing a Factor Xa site between the MBP and paramyosin domains.

Molecular Weight: 70.2 kDa.

Concentration and Shipping: 5 mg/ml. Supplied in 20 mM Tris-HCl (pH 7.2), 0.2 M NaCl, 10 mM b-mercaptoethanol, 50% glycerol and 1 mM sodium azide. Store at .20°C.

E8051S	100 µg	$50 (USA)

MBP2* Protein:

Description: MBP2* is wild type MBP plus the amino acids encoded by the pMAL-c2 polylinker. It is purified from cells bearing a derivative of pMAL-c2 with a linker containing a stop codon inserted in the Xmn I site.

Molecular Weight: 42.5 kDa.

Concentration and Shipping: 4–8 mg/ml. Supplied in 20 mM Tris-HCl (pH 7.2), 0.2 M NaCl, 1 mM EDTA, and 50% glycerol. Store at –20°C.

E8044S	1.0 mg	$50 (USA)
E8044L	5.0 mg	$200 (USA)

malE and M13/pUC sequencing primer: (*not supplied with the system*)

malE Primer: Used for sequencing downstream from the malE gene across the polylinker.
5'(dGGTCGTCAGACTGTCGATGAAGCC)3'

M13/pUC sequencing primer: Used for sequencing upstream from the *lacZ*a gene across the polylinker.
5'(dCGCCAGGGTTTTCCCAGTCACGAC)3'

S1237S	malE Primer	0.5 A_{260} units	$95 (USA)
S1224S	M13/pUC Sequencing Primer	0.5 A_{260} units	$95 (USA)

M13KO7 Helper Phage: (*not supplied with the system*)

N0315S	M13KO7 Helper Phage	1.8 ml	$55 (USA)

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NEW ENGLAND BioLabs Inc.

Positive selection vectors to generate fused genes for the expression of his-tagged proteins.

Van Reeth T, Dreze PL, Szpirer J, Szpirer C, Gabant P.

Universite Libre de Bruxelles, Rhode-Saint-Genese, Belgium.

Epitope tagging simplifies detection, characterization and purification of protei Gene fusion to combine the coding region of a well-characterized epitope with coding region for a protein of interest generally requires several subcloning ste Alternatively, a PCR strategy can be used to generate such a chimeric gene. In addition to its simplicity, this approach allows one to limit the size of the multi cloning sites present in conventional expression vectors, thus reducing the introduction of artifactual amino-acid sequences into the fused protein. In this communication, we describe new vectors that allow PCR cloning and selection chimeric genes coding for N- or C-terminal His-tagged proteins. These vectors based on the control of cell death CcdB direct selection technology and are well adapted to the cloning of blunt-ended PCR products that were generated by usir thermostable polymerases that provide proofreading activity.

PMID: 9821593 [PubMed - indexed for MEDLINE]

1: Anal Biochem. 1999 Apr 10;269(1):10-6. Related Articles, Lir

Influence of the amino acid residue downstream of (Asp)4Lys on enterokinase cleavage of a fusion protein.

Hosfield T, Lu Q.

Stratagene Cloning Systems, Inc., La Jolla, California 92037, USA.
tanya_hosfield@stratagene.com

We have studied the cleavage efficiency of the protease enterokinase (EK) usin the novel vector pESP4. pESP4 is a yeast expression vector equipped with ligation-independent cloning sites, a GST purification tag, and a FLAG epitope tag. EK is used to cleave the FLAG and GST tags leaving the protein of interest without any extraneously added amino acids. We have found that EK is relative permissive of the amino acid residue downstream of the recognition sequence (t P'1 position). This makes EK an ideal choice to use as a protease to cleave any protein of interest cloned within the pESP4 yeast expression vector.

PMID: 10094769 [PubMed - indexed for MEDLINE]

1: J Biotechnol. 1998 Jun 30;62(2):143-51. Related Articles, Lir

High-level production of human growth hormone in Escherichia coli by a simple recombinant process.

Shin NK, Kim DY, Shin CS, Hong MS, Lee J, Shin HC.

Laboratory of Protein Engineering, Hanhyo Institute of Technology, Yusong-ku Taejon, South Korea.

Procedures have been devised for producing in Escherichia coli high yields of purified recombinant human growth hormone (hGH), by utilizing N-terminal pentapeptide sequence of human tumor necrosis factor-alpha, histidine tag and enterokinase cleavage site as a fusion partner. The fusion protein was produced a soluble protein at the beginning of gene expression, but progressively became insoluble in Escherichia coli cytoplasm. The insoluble protein was solubilized b simple alkaline pH shift and purified to near homogeneity by Ni(2+)-chelated affinity chromatography. Following specific enterokinase cleavage, the recombinant hGH was purified by one-step anion exchange chromatography. Tl ease and speed of this recombinant process, as well as the high productivity, makes it adaptable to the large-scale production of hGH. Moreover, the highly efficient fusion partner could be applied to the production of other therapeutical important proteins.

PMID: 9706704 [PubMed - indexed for MEDLINE]

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(FILE 'HOME' ENTERED AT 19:09:17 ON 30 OCT 2003)

INDEX 'ADISCTI, ADISINSIGHT, ADISNEWS, AGRICOLA, ANABSTR, AQUASCI, BIOBUSINESS, BIOCOMMERCE, BIOSIS, BIOTECHABS, BIOTECHDS, BIOTECHNO, CABA, CANCERLIT, CAPLUS, CEABA-VTB, CEN, CIN, CONFSCI, CROPB, CROPU, DISSABS, DDFB, DDFU, DGENE, DRUGB, DRUGLAUNCH, ...' ENTERED AT 19:10:57 ON 30 OCT 2003

SEA TAG? (S)(VECTO? OR PLASMI?) (S) CLEAV? (S) SECRET?

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4* FILE FEDRIP
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10 FILE GENBANK
34 FILE IFIPAT
13 FILE LIFESCI
7 FILE PASCAL
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2 FILE USPAT2
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15 FILE WPINDEX
1 FILE NLDB

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L7 10 S L6 AND (NEUROS? OR KALLIKR?)